Chapter 7

Osteoporosis and Nutrition — Nutrition, Anthropometry and Bone Mineral Density in Women

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1. Introduction

Osteoporosis affects millions of people all around the world and it is the most common metabolic bone disease, characterized by low bone mass, disrupted bone micro architecture and increased bone brittleness [1].

Interaction of numerous factors, such as: genetic, medical, anthropometric, pharmacological, lifestyle and nutrition, lead to loss of bone mass and to increased risk for the osteoporotic fractures in female [2].

The most of the studies which have explored the effect of calcium on bone mass in females, demonstrated that high calcium intake is related to greater bone mass, compared to smaller bone mass in respondents who had less dietary calcium intake [3]. Besides calcium, sufficient dietary intake of other micronutrients, such as: zinc, magnesium, potassium, dietary fibers as well as vitamin C are believed to have favorable effect on the bone metabolism too [4].

The study of osteoporotic fractures reports that higher intake of animal proteins compared to vegetable proteins is associated to increased risk of loss of the bone mass and occurrence of the osteoporotic fractures [5]. High protein and sodium intake increases calcium excretion in urine, which increases the need for dietary calcium. It has been also found that a high dietary total protein intake, increases production of endogenous acid, which results in accelerated bone resorption and reduced bone formation. This is especially expressed in diets high in animal proteins [6]. It is believed that unfavorable effect of the animal proteins on the bone metabolism can be repaired by higher fruit and vegetable intake [7].

Recent researches indicate a risk of excessive fat intake, which leads to metabolic bone disorders. High fat intake is considered to be a risk factor for osteoporosis, because it reduces the calcium absorption, since calcium forms insoluble compounds with fatty acids [7].

The aim of this study was to quantify the intake of trace elements in fruit and vegetable: zinc, magnesium, potassium and dietary fats as well as fat derivatives intake in examinees and to explore their relation to the bone mass. The aim was to examine the extent to which these nutritional parameters are predictors of values of bone mineral density.

2. Patients and methods

2.1. Subjects

The study population consisted of women with sedentary occupations in age ranged from 40 to 67 years. Women are inhabitants of the down town Rijeka, Croatia. Exclusion criteria for further participation in the survey were: smoking and any medical therapy which can alter bone metabolism, including food supplements with added calcium. Dietary habits, anthropometric characteristics, serum concentration of the biochemical markers and values of the bone densitometry parameters were comprised by this study. 200 women were included in this investigation, of which 120 menopausal women constituted experimental group, and 80 fertile women represents the control group.

2.2. Dietary intakes

Participants completed an anonymous, encrypted questionnaire, conducted in accordance with ethical and bioethical principles and their privacy and protection of confidential information was ensured.

For the assessment of dietary habits and the average daily energy and nutrients intake, we used data obtained from semi- quantitative Food Frequency Questionnaire- sq-FFQ, the main method for collecting data about a foodstuff choice, as well as the type and quantity of food intake in the study population. This method of identifying the dietary habits is a questionnaire validated by the Department of Nutrition, Harvard School of Public Health [8], from which are obtained informations about daily intake of energy and nutrients. Women were asked to note the frequency and the quantity of offered food items. The amount of each food item was offered as one portion and declared as small, medium and large. This method quantified the values of nutritional parameters that are essential for bone health, such as: calcium, phosphorus, vitamin D, proteins, zinc, magnesium, potassium, dietary fibers and vitamin C. We also determined a total fat intake and the emphasis was placed on the intake of total fat, saturated fatty acids, monounsaturated fatty acids and polyunsaturated fatty acids. The nutritive and energy values of each food noted were calculated using the composition tables of raw and cooked food [9].

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2.3. Anthropometric, biochemical and bone mineral status measurements

Bodyweight was measured on a portable electronic scale (SECA, Hamburg, Germany), with accuracy of ± 0,1 kg. Body height was measured on a portable stadiometer, which is a part of a specified scale (SECA, Hamburg, Germany), to the nearest ± 0,5 cm. Body mass index was calculated as bodyweight divided by body height squared, BMI (kg/m²) [10].

Biochemical indicator of bone resorption, deoxypyridinolin (DPD) and biochemical indicator of bone formation, bone alkaline phosphatase (ALP) and vitamin D were determined from urine and blood of the respondents by immune-enzymatic method (Enzyme Linked Immunosorbent Assay, ELISA), according to manufacturer's protocol [11,12,13].

Bone density in the anterior-posterior images of the spine and hip was measured using the device for bone densitometry (Hologic, Bedford, MA, USA). The obtained values were quantified according to the following parameters: bone mineral content (BMC, mg), bone mineral density (BMD, mg/cm²) and T-score (represents a deviation from the BMD measured values of peak bone mass of young people expressed in standard deviations) and Z-score (deviation of the measured values BMD of the average bone mass of persons of the same age, expressed in standard deviations) [14].

2.4. Statistical analysis

Statistical analysis of data was performing by using Statistica for Windows, release 9.1 (Stasoft, INC, Tulsa, USA). Normality of distribution for the data interval scale (quantitative data), was tested using the Kolmogorov- Smirinov test. The results were shown as arithmetic mean and standard deviation. Results were distributed normally and in the analytical statistics, one-way analysis of variance (one-way ANOVA) was used. To determine the significance of the contribution of the percentage of nutrients on the metabolic bone status, multiple regression analysis was used. All statistical values were considered significant at the level $P<0,05$ [15].

2.5. Results

Age, anthropometric characteristics, values of the bone densitometry parameters and concentrations of the bone remodeling markers are presented in Table 1. Women of generative age are significantly taller than women in menopause ($P=0,01$) and have significantly higher body weight than women in menopause ($P<0,001$). The average value of Body Mass Index (BMI) was 27 kg/m².

Subjects of generative age have significantly higher values of BMD and BMC of the spine ($P<0,001$, $P<0,001$), as well as the values of T-score and Z-score ($P<0,001$, $P<0,001$), than menopausal women, respectively. Values of BMD and BMC of the hip ($P<0,001$, $P<0,001$) and the value of T-score ($P<0,001$) were also significantly higher in women of generative age.

Regarding the bone remodeling markers, significantly lower values of DPD ($P<0,001$) and bone ALP ($P=0,004$) were found in fertile women compared to menopausal women.

Parameters	Fertile women (n =80)	Menopausal women (n = 120)	Total (n = 200)	P-value
Age	47,6 ±4,1	59,9 ± 5,1	54,9 ± 7,7	<0,001*
Body height (cm)	74,0 ± 6,4	71,7 ± 13,3	72,6 ± 11,1	0,001*
Body weight (kg)	166,8 ± 0,05	161,9 ± 0,06	163,8 ± 0,06	<0,001*
BMI (kg/m^2)	26,6 ± 2,3	27,3 ± 4,7	27,0 ±3,9	0,210
BMD LS (g/cm^2)	1,074 ± 0,1	0,897 ± 0,1	0,968 ± 0,2	<0,001*
BMC LS (g)	67,49 ± 9,4	52,84 ± 9,7	58,70 ± 11,9	<0,001*
T-score	0,400 ± 1,3	-1,325 ± 1,3	-0,635 ± 1,9	<0,001*
Z-score	0,835 ± 1,3	0,033 ± 1,4	0,354 ± 1,4	<0,001*
BMD LH (g/cm^2)	0,944 ± 0,1	0,860 ± 0,1	0,893 ± 0,1	<0,001*
BMC LH (g)	37,29 ± 5,9	30,83 ± 5,5	33,41 ± 6,5	<0,001*
T-score	0,122 ± 0,8	-0,647 ± 1,1	-0,339 ± 1,1	<0,001*
Z-score	0,453 ± 0,9	0,298 ± 1,0	0,360 ± 1,0	0,269
DPD (nmol/l)	5,26 ± 1,4	6,85 ± 2,5	6,22 ± 2,2	<0,001*
ALP (ng/ml)	22,77 ± 8,1	26,0 ± 7,4	24,71 ± 7,8	0,004*
Vitamin D (nmol/l)	62,03 ± 25,8	68,90 ± 29,1	66,16 ± 27,9	0,09

* statistical significance on level P < 0,05

LS – lumbar spine

LH – left hip

Table 1. Age, anthropometry, bone densitometry parameters, bone remodeling markers and vitamin D ($\bar{X} \pm SD$)

Dietary habits of the study participants are presented in the Table 2. One-way analysis of variance (ANOVA) showed that women of generative age have significantly higher average daily intake of vitamin D, vitamin C, potassium, magnesium and zinc, while menopausal women have significantly higher average daily phosphorus intake (P<0,001).

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Parameters		Fertile women (n =80)	Menopausal women (n = 120)	Total (n = 200)	P- values
Energetic food equivalent	kcal	2851,91 ± 1034,4	2448,65 ± 716,37	2609,95 ± 877,9	<0,001*
	kJ	11932,38 ± 4327,8	10,245 ± 2997,3	10920,04 ± 3673,5	<0,001*
Proteins (total) (g)		90,76 ± 65,4	60,52 ± 28,9	75,64 ± 50,7	<0,001*
Proteins (vegetable) (g)		21,95 ± 22,5	19,70 ± 9,2	20,71 ±16,7	<0,001*
Proteins (animal) (g)		67,88 ± 45,5	40,20 ± 24,7	54,04 ± 37,0	<0,001*
Total fat (g)		101,74 ± 63,9	74,76 ± 45,37	87,30 ± 56,3	<0,001*
Saturated fatty acids (g)		45,35 ± 28,3	31,41 ± 22,6	38,38 ± 25,8	<0,001*
Monounsaturated fatty acids (g)		34,40 ± 25,32	26,37 ± 18,2	30,39 ± 22,1	<0,001*
Polyunsaturated fatty acids (g)		20,89 ± 13,05	16,98 ± 8,2	18,94 ± 11,5	<0,001*
Carbohydrates (g)		180,16 ± 215,9	152,05 ± 90,2	166,10 ± 163,8	<0,001*
Vegetable fibers (g)		26,72 ± 24,1	14,27 ± 11,5	20,50 ± 18,7	<0,001*
Vitamin D (μg)		9,91 ± 5,1	6,32 ± 7,6	7,76 ± 6,9	<0,001*
Vitamin C (mg)		131,62 ± 111,1	118,36 ± 112,0	123,66 ± 166,8	<0,001*
Calcium (mg)		953,91 ± 316,32	918,79 ± 232,0	932,74 ± 268,7	0,366
Phosphorus (mg)		1012,23 ± 315,23	1132,21 ± 235,25	1072,22 ± 235,2	<0,001*
Potassium (mg)		6441,99 ± 3231,4	4453,70 ± 1362,0	5294,02 ± 2482,4	<0,001*
Magnesium (mg)		546,16 ± 245,9	404,70 ± 136,1	461,28 ± 199,8	<0,001*
Zinc (mg)		17,38 ± 8,0	13,02 ± 4,2	14,77 ± 6,4	<0,001*

* statistical significance on the level P < 0,05

Table 2. The average daily nutrient intake in fertile and in menopausal women ($\bar{X}$ ± SD)

DXA parameters which were extracted by ROC analyses as excellent predictors of bone metabolism were included in multiple regression analyses. Those include: LS BMC, LS BMD, LS T-score, LS Z-score, LH BMC, LH T-score.

The results of multiple regression analysis by which are defined total shares and significance of contributions of menstrual status, age, anthropometry and nutrition on the bone densitometry parameters. The largest total share of contributions to all the bone densitometry parameters was observed for menstrual status and diet (Table 3).

Parameters	Menstrual status	Age	Anthropometry	Nutrition
	Share of contributions (%)	Share of contributions (%)	Share of contributions (%)	Share of contributions (%)
LS BMC	40,1	8,4	8,5	44,8
LS BMD	29,6	3,3	16,6	12,7
LS T-score	29,6	6,1	19,3	27,4
LS Z-score	20,8	7,2	13,0	12,7
LH BMC	27,8	12,0	24,2	23,2
LH T-score	24,5	0,5	27,0	18,6

LS – lumbar spine

LH – left hip

Table 3. Total shares of contributions of menstrual status, age, anthropometry and nutrition on DXA (%)

Parameters	LS BMC		LS BMD		LS T-score		LS Z-score		LH BMC		LH T-score	
	ß	P	ß	P	ß	P	ß	P	ß	P	ß	P
Menstrual status	-0,632	<0,001*	-0,560	<0,001*	-0,632	<0,001*	-0,529	<0,001*	-0,568	<0,001*	-0,389	<0,001*
Age	-0,156	0,057	-0,097	0,251	-0,156	0,057	-0,365	0,001*	-0,210	0,010*	-0,017	0,831
Nutrition												
Energy	0,346	0,014*	0,238	0,150	0,667	0,016*	0,627	0,002*	0,400	0,048*	0,063	0,529
Proteins (g)	0,008	0,580	0,010	0,445	0,081	0,481	0,004	0,978	0,349	<0,001*	0,786	0,082
Total fat (g)	-0,273	0,036*	-0,103	0,386	-0,098	0,016*	-0,191	0,127	-0,427	0,016*	-0,500	0,054
Saturated fatty acids (g)	-0,234	0,810	-0,141	0,166	0,148	0,005*	-0,016	0,332	-0,414	0,053	-0,259	0,244
Monounsaturated fatty acids (g)	0,079	0,551	0,029	0,875	0,144	0,019*	0,031	0,145	0,328	0,182	0,258	0,312
Polyunsaturated fatty acids (g)	0,086	0,546	0,044	0,782	0,209	0,049*	0,013	0,104	0,310	0,069	0,329	0,063
Calcium (mg)	2,045	0,256	0,538	0,168	0,007	0,145	-0,219	0,032*	0,034	0,626	0,023	0,748
Potassium (mg)	0,211	0,004*	0,536	0,006*	0,213	0,005*	0,103	0,004*	0,089	0,001*	0,750	0,414
Phosphorus (mg)	-0,623	0,277	-0,078	0,053	0,139	0,031*	0,071	0,033*	-0,078	0,851	-0,234	0,588
Magnesium (mg)	0,073	0,002*	0,054	0,053	0,133	0,036*	0,031	0,010*	0,157	0,002*	0,422	0,187

*statistical significance on level P < 0,05

β – regression coefficient

LS – lumbar spine

LH – left hip

Table 4. Statistically significant interactions of predictors (menstrual status, age, and nutrients) to categorical variables (LS BMC, LS BMD, LS T-score, LS Z-score, LH BMC, LH T-score) are shown

Menstrual status, age, total fat, saturated fatty acids, are inversely proportional to the values of the bone densitometry parameters, while energy, proteins, monounsaturated fatty acids, polyunsaturated fatty acids, calcium, potassium and magnesium are exactly proportional to the values of the bone densitometry parameters.

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Parameters	Milkand milk products		Fish		Vegetables		Fruit	
LS BMD	β	P	β	P	β	P	β	P
	0,008	0,169	0,031	0,677	0,034	0,742	0,027	0,733
LS BMC	0,109	0,128	0,171	0,014*	0,006	0,346	0,016	0,645
LS T-score	0,077	0,301	0,043	0,577	0,089	0,150	0,047	0,556
LS Z-score	0,105	0,163	0,008	0,024*	0,127	0,135	0,092	0,256
LH BMC	0,039	0,598	0,086	0,005*	0,214	0,002*	0,156	0,050
LH T-score	0,178	0,015*	0,135	<0,001*	0,178	0,004*	0,107	0,178

*statistical significance on level P < 0,05

β – regression coefficient

LS – lumbar spine

LH – left hip

Table 5. Interactions of predictors (milk and milk products, fish, vegetables and fruit) to categorical variables (LS BMD, LS BMC, LS T-score, LS Z-score, LH BMC, LH T-score) are shown

Milk and milk products, fish, vegetables and fruit are exactly proportional to the bone densitometry parameters.

Parameters	LS BMC	LS BMD	LS T-score	LS Z-score	LH BMC	LH T-score
	share of contribution (%)	share of contribution (%)	share of contribution (%)	share of contribution (%)	share of contribution (%)	share of contribution (%)
Menstrual status	40,1	29,6	29,6	20,8	27,8	24,5
Age	8,4	3,3	6,1	7,2	12,0	0,5
Energy	2,7	0,5	2,8	3,2	1,4	0,2
Proteins (g)	0,2	0,1	0,2	0	5,8	4,8
Total fat (g)	5,8	0,9	2,4	1,8	1,5	2,4
Saturated fatty acids (g)	0,5	1,6	3,3	0,2	1,7	0,7
Monounsaturated fatty acids (g)	1,4	0,2	2,2	0,3	1,5	0,7
Polyunsaturated fatty acids (g)	1,6	0,5	1,6	0,1	0,2	0,6
Calcium (mg)	0,9	0,7	1,0	1,8	0,2	0
Potassium (mg)	8,8	5,9	3,4	4,1	7,3	5,1
Phosphorus (mg)	5,3	0,6	3,5	4,5	0	0,9
Magnesium (mg)	10,1	0,2	3,1	2,4	2,8	0,8

Table 6. Total shares of contributions of menstrual status, age and nutrition on DXA (%)

3. Discussion

It is considered that menopausal women have the highest risk of osteoporotic fractures and the incidence of osteoporosis in this group is increased by 25% compared to fertile women [3]. Furthermore, the frequency of osteoporotic vertebral and hip fractures in both genders increases exponentially with age [1]. Results obtained by this study coincide with the majority of results of similar studies, showing that menopausal women have significantly lower values of the bone densitometry parameters [17,18,19]. The latter is additionally confirmed by our finding of inversely proportional relationship between menopause and bone mineral density parameters. Our result of inversely proportional relationship between age and bone mineral density parameters, corresponds to a study conducted on 450 000 participants from Sweden [20].

Bone remodeling markers provide information about the dynamic state of bone metabolism, and those are very useful tools to predict early changes in the bone metabolism. Along with the bone densitometry, bone remodeling markers are needy in diagnosis and follow up of diseases of the bone mass deficit [21]. We have demonstrated that women of generative age have significantly lower values of DPD ($P<0,001$) than women in menopause. This is consistent to increased excretion of DPD in postmenopausal women [22]. Menopausal women had significantly higher levels of bone ALP ($P=0,004$) compared to women in generative age. The latter could be explained by the fact that high serum concentration of bone formation markers is associated with greater bone loss [23]. Average concentration of vitamin D in all study participants amounted 66,16 nmol/l, out of which average value in fertile women amounted 62,03 nmol/l, and 68,9 nmol/l in menopausal women (Table 2). Similar values of vitamin D concentrations were observed in a study of postmenopausal women of nine European countries [24]. By adding our results of low vitamin D concentrations to similar results published by Kraljević et al. [25] and by Žerjavić et al. [26], we can summarize that some action should be done by Croatian Health Care system, such as food-based strategies, to prevent vitamin D deficiency in Croatia.

Nutrition has a unique role in processes of growth and modeling of the human skeleton, as well as in maintaining the peak bone mass in adulthood [2]. The most of the studies are focused on dietary calcium intake [19], some of the researchers have analyzed the impact of some other nutrients such as proteins, carbohydrates, fat and energy intake [27], and only a few studies have explored the influence of vitamins and minerals on bone mass [7]. Calcium in a form of the calcium phosphate or calcium carbonate is the major mineral constituent of the bone tissue. High calcium intake, within the normal diet, does not protect against fractures, but low calcium intake represents a risk factor for the osteoporosis [19].

By means of the multiple regression analysis we have determined that the greatest contribution of the anthropometry is to values of the BMC and the T-score of the left hip

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(Table 3). A positive relationship between increased body weight or body mass index (BMI) to bone mass has been already reported [28,29]. Some authors have considered that increased body weight can improve bone mass, by stimulation of the osteoblast differentiation. Body weight increase in postmenopausal period is correlated to increased number of adipocytes. Adipocytes are an important source of estrogen, a hormone which stimulates bone formation [30]. The opposite theories to afore mentioned have been reported too [31] and amongst those is a research of Kroke et al. [32], who did not find strong influence of anthropometric parameters on bone mineral density neither in women of generative age, nor in postmenopausal women.

It has been proposed that high energy intake, leads to body weight increase and finally to increased values of the bone mineral density [33,34]. Similarly to results obtained by Kumar et al. [35], our results indicate that daily energy intake is exactly proportional to the values of the bone densitometry parameters (Table 4).

Total daily protein intake is directly proportional to the values of the bone densitometry parameters, which was significant for the LH BMC ($P<0,001$) (Table 4). Such result is in agreement to results obtained by Misra et al. [36]. These researchers have documented a positive correlation between total protein intake and increased bone mineral density. Further protein analysis revealed a positive influence of proteins of vegetable origin and negative influence of proteins of animal origin on the bone mass. SOF study (Study of osteoporotic fractures) found that women with increased animal proteins intake have low values of the bone mineral density and increased risk of osteoporotic fractures [5].

Total fat and saturated fatty acids are inversely proportional to DXA parameters, while monounsaturated and polyunsaturated fatty acids are exactly proportional to DXA parameters (Table 4). Greater shares of contribution of all four types of fat were found for DXA parameters of the lumbar spine than of the left hip (Table 6). Significance was observed for the T-score of the lumbar spine (Table 4). Corwin et al., conducted the survey on menopausal women, and found a negative correlation between total fat intake and bone mineral density [37]. Another research of Hogstroma et al. corresponds to our results since they have also found positive correlation between monounsaturated fatty acids intake and bone mineral density [38].

Regarding daily calcium intake, total shares of calcium contribution are greater in lumbar spine than in the left hip. Interestingly calcium does not contribute at all to the values of the LH T-score (Table 6). Calcium is directly proportional to the values of DXA parameters, but the only statistical significance relates to LS Z-score (Table 4). Similarly was found in the study of H. F. Saadi et al., where dietary calcium intake of fertile women and postmenopausal women is positively, but not significantly correlated to bone mineral density [39]. Average daily calcium intake amounted 932,74 g, which are adequate amounts of dietary calcium according to existing recommendations in Croatia (Table 2).

Daily intake of the minerals magnesium and potassium has the greatest contribution of all the given minerals to DXA parameters (Table 6). Both of the minerals are directly proportional to DXA parameters (Table 4). Studies published so far argue that sufficient magnesium and potassium intake is related to increased bone mineral density [40,41], or as contrast opinions state, to reduced bone mass and increased risk of wrist fracture [43]. Magnesium is important in processes of bone mineralization, and potassium has important role in systemic acid-base (pH) homeostasis. Potassium salts neutralize bone-depleting metabolic acids, and therefore conditions that require drain of alkalizing compounds from bone lead to loss of bone tissue. Positive influence of potassium on bone health has been reported [3,4,40,45].

Zinc is a cofactor for alkaline phosphatase, an enzyme, necessary for bone mineralization. Low concentration of the zinc in serum and its increased secretion in urine is associated with osteoporosis [44]. Considering that calcification of the bone is reduced with insufficient zinc intake, we analyzed influence of the dietary zinc on bone health. Results revealed that daily zinc intake is directly proportional to DXA parameters, but the influence was not significant (data not shown).

Analyzing the impact of fruit and vegetables on DXA parameters in women, we have found that vegetables are directly proportional to the parameters of the left hip, which is statistically significant (table 5). Similar results were obtained by New et al. [3] who have that bone mineral density in premenopausal women was positively related to fruit and vegetable intake, as well as to magnesium, calcium, zinc and plant fibers. Similarly, Tucker et al. have found better bone mass in women who consumed more fruits, vegetables, potassium and magnesium [41]. However, other study which has included postmenopausal women, showed no relationship between bone mass to fruit or vegetable intake [7,46].

Regarding the mechanisms fruit and vegetables influence bone metabolism, it is important to mention that these nutrients create an alkaline environment and therefore reduce urinary calcium excretion. Besides, fruits and vegetables are rich in vitamins with antioxidant properties such as vitamin C and beta-carotene. Vegetables are an important source of vitamin K, which also has a role in the mineralization of bone since it induces carboxylation of osteocalcin [40].

4. Conclusion

Analyses of the impact of age, anthropometric parameters, menstrual status and nutrition on the bone status, represents the age and menstrual status as predictors with the highest influence on the bone mineral density in women.

Fruits and vegetables have pleiotropic effects on bone metabolism, which include: alkalinity, antioxidant properties of vitamins and as it was determined by this study, beneficial influence of minerals magnesium, potassium and zinc.

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